

FIG. 1

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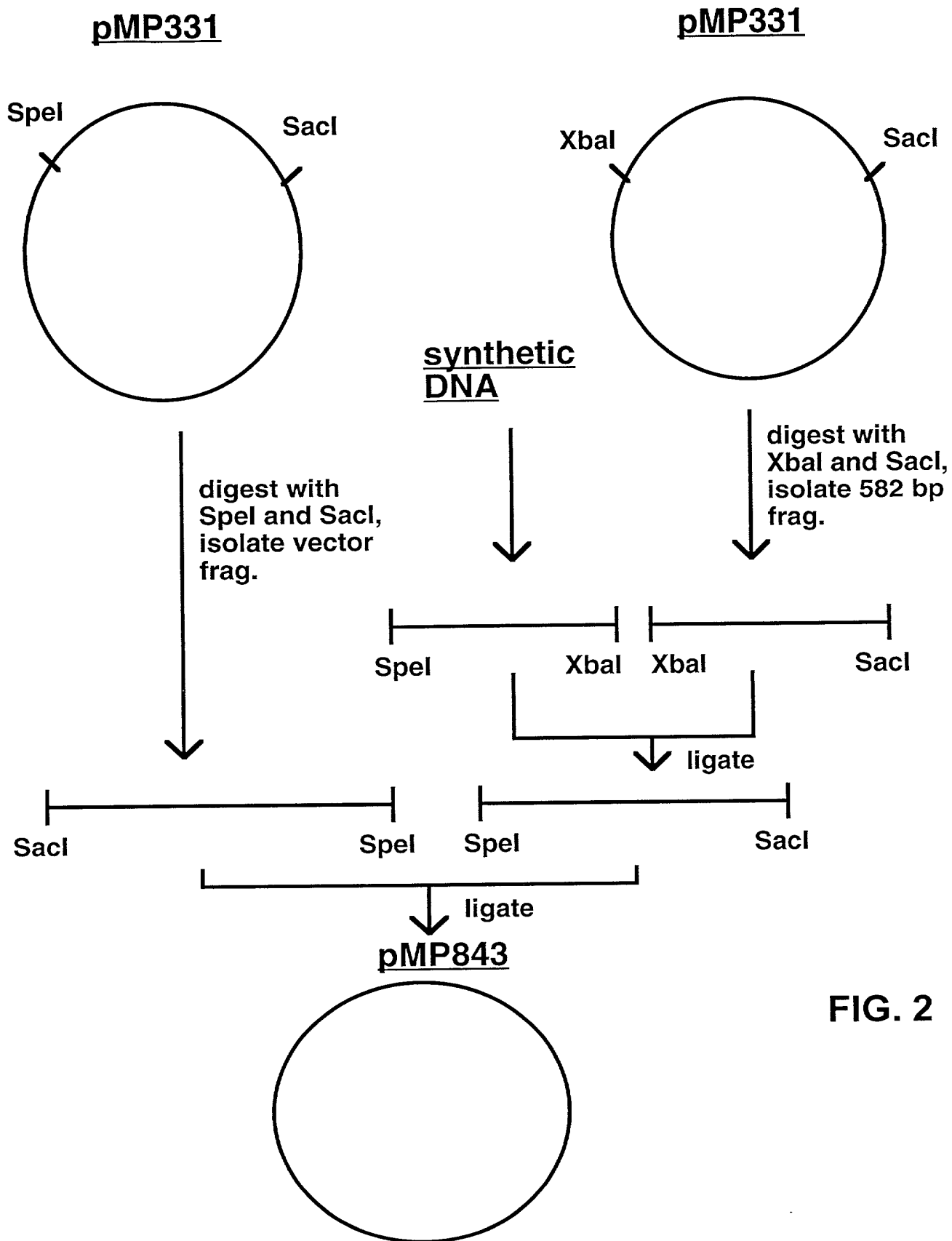


FIG. 2

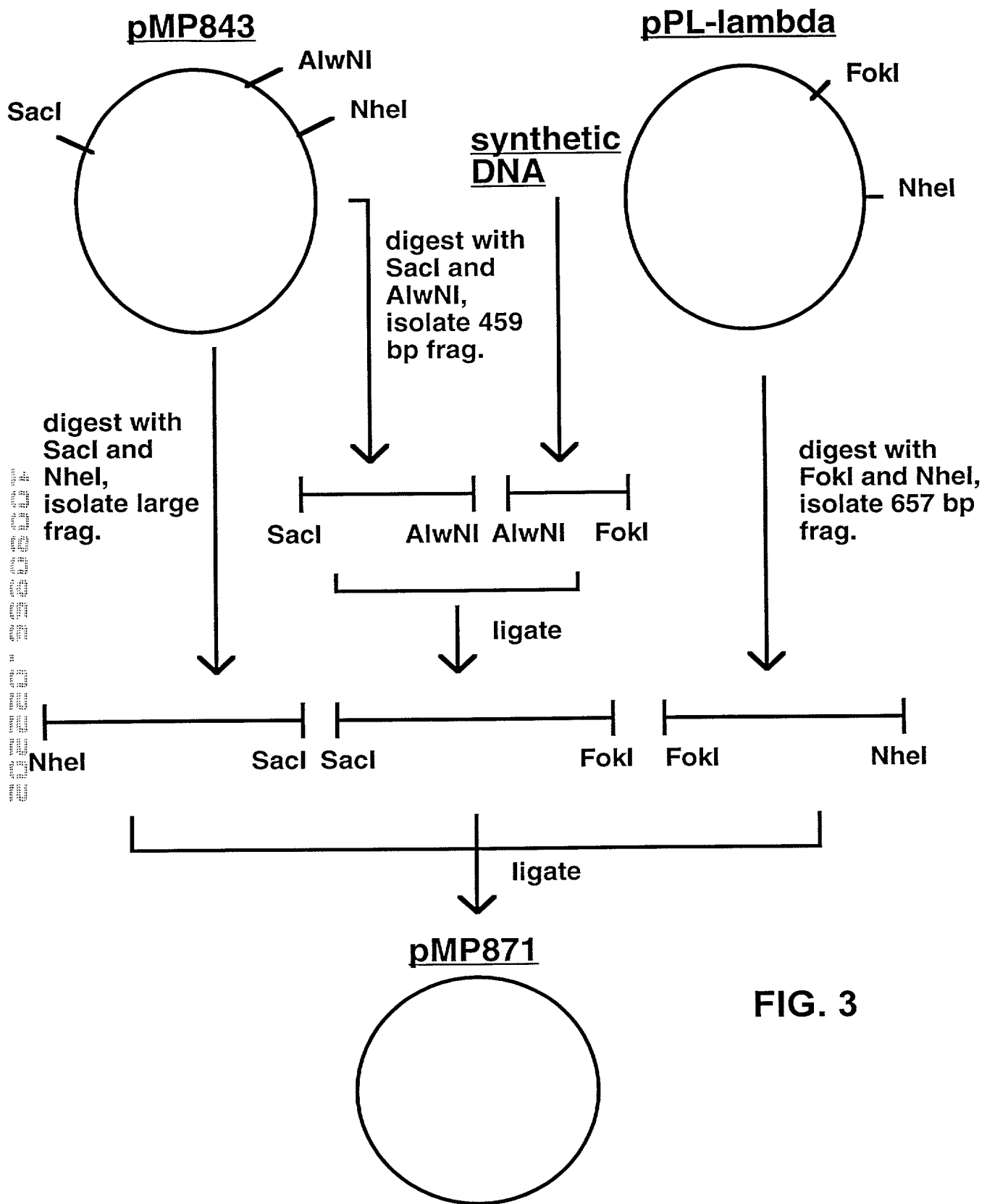
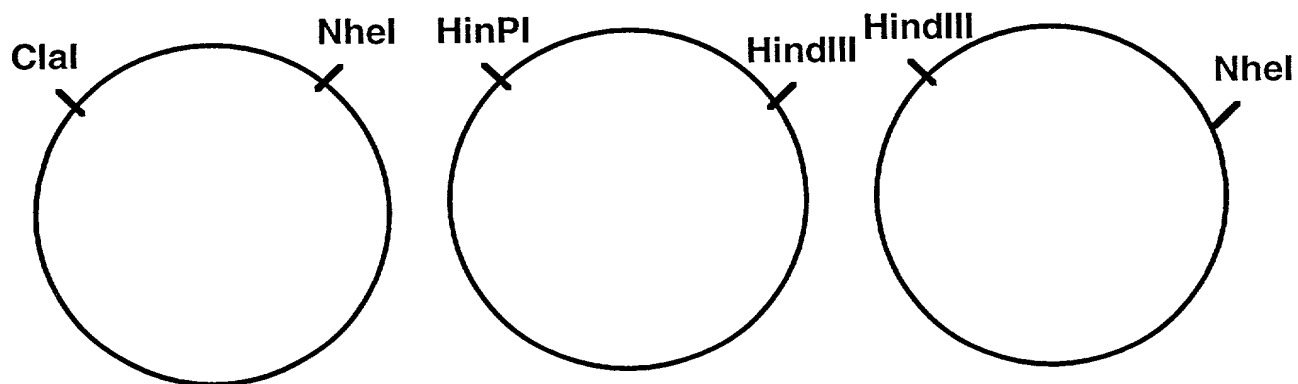


FIG. 3

pMP843

pKMTacII

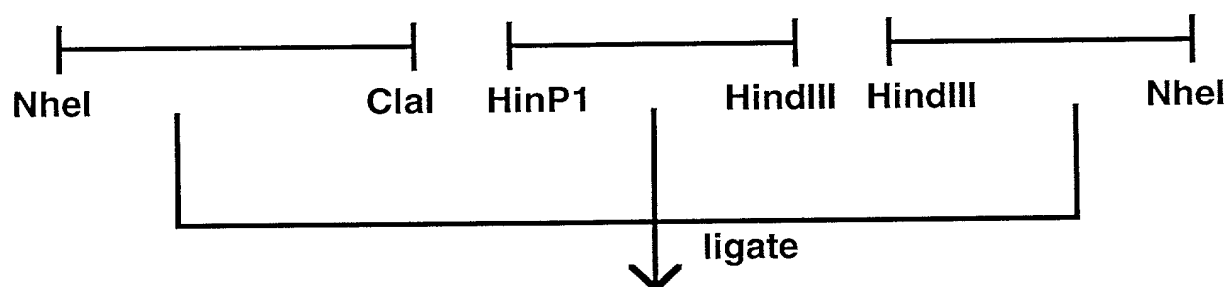
pMP871



digest with *Clal* and *NheI*,
isolate large frag.

digest with *HinPI* and *HindIII*,
isolate 100 bp frag.

digest with *HindIII* and *NheI*, isolate
684 bp frag.



pMP931

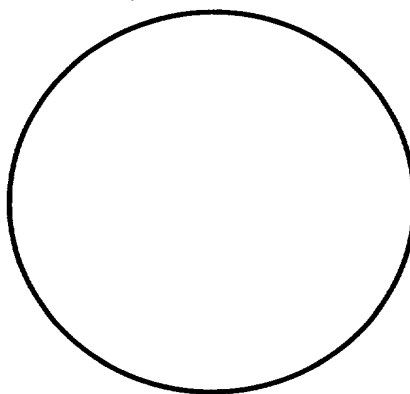
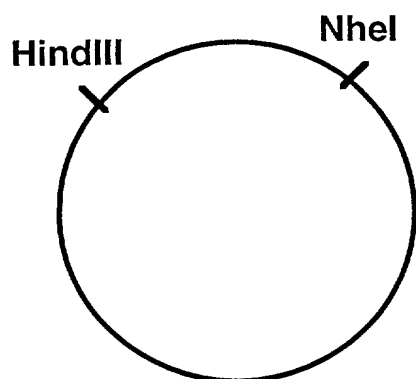
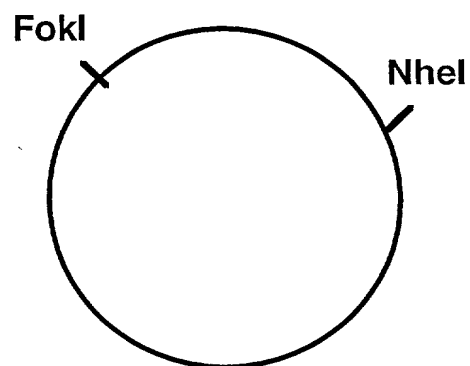


FIG. 4

pMP931



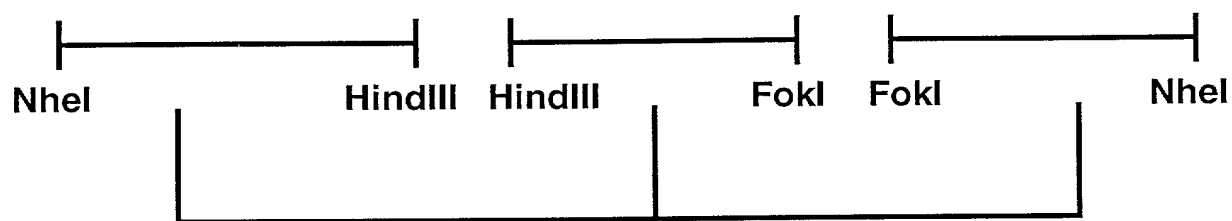
pPL-lambda



synthetic
DNA

digest with
HindIII and
NheI, isolate
large frag.

digest with
FokI and NheI,
isolate 657 bp
frag.



ligate

pMP945

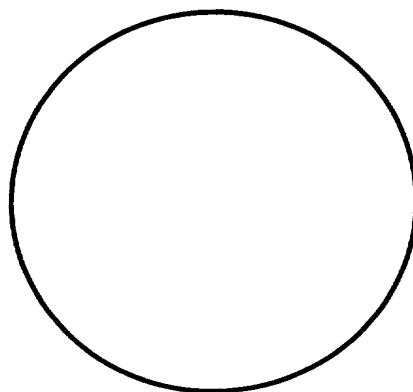
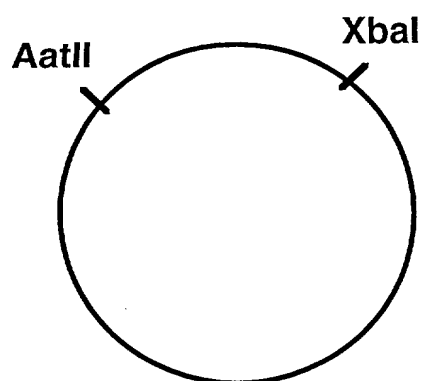


FIG. 5

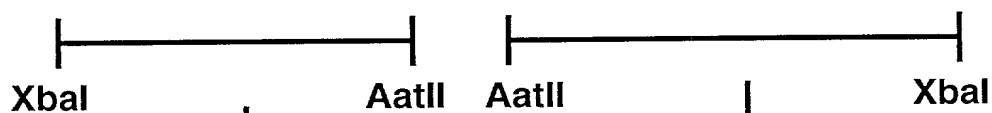
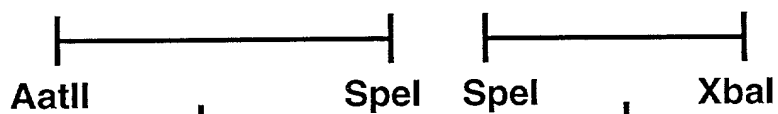
pMP931



digest with AatII
and XbaI, isolate
large frag.

digest with AatII
and SpeI, isolate
322 bp frag.

**synthetic
DNA**



pMP951

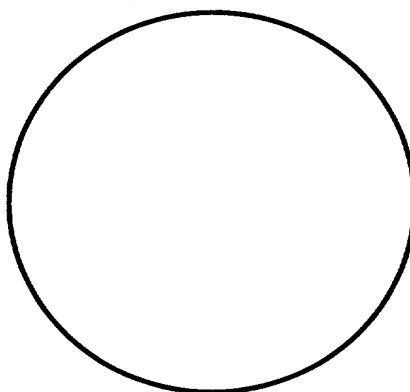
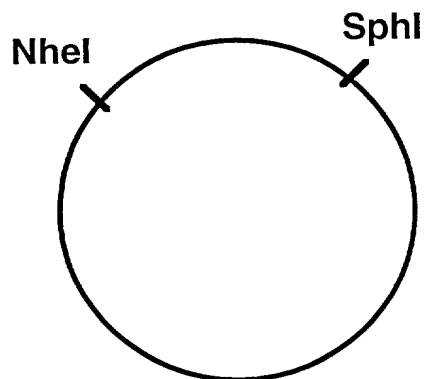


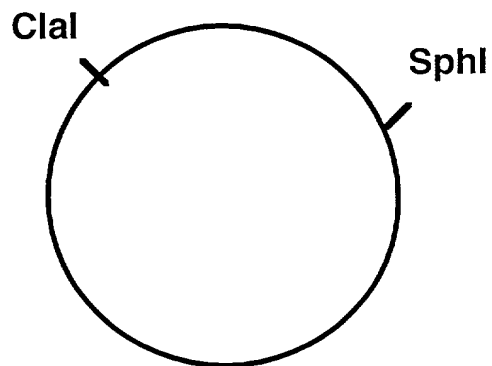
FIG. 6

pMP951

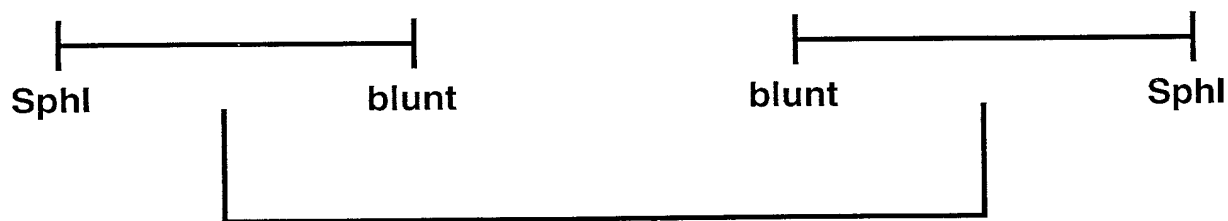


digest with NheI,
fill in with DNA
polymerase
(Klenow)
second cut with SphI,
isolate large frag.

pST141



digest with ClaI
fill in with DNA
polymerase
(Klenow),
second cut with
SphI, isolate
440 bp frag.



ligate

pMP982

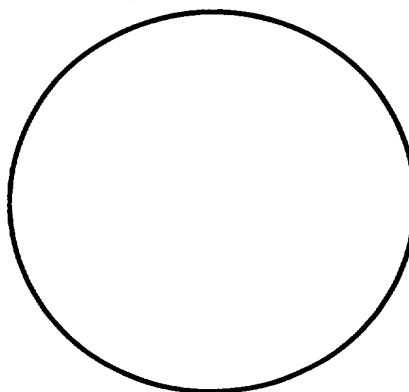
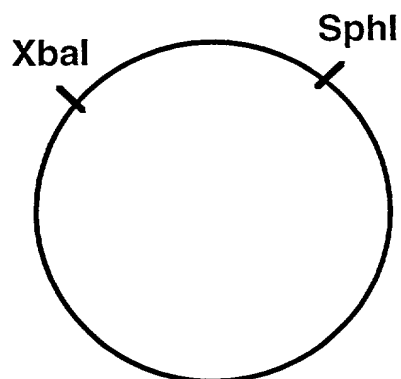


FIG. 7

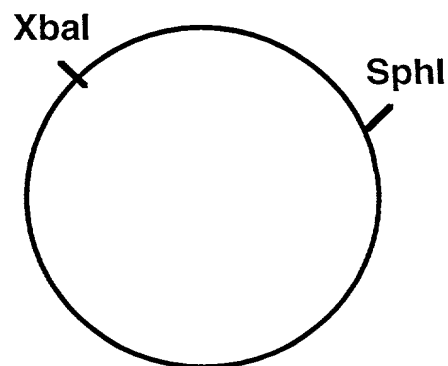
pST182



digest with XbaI
and SphI, isolate
large frag.



pMP331



digest with XbaI
and SphI, isolate
1791 bp frag.



ligate

pMP1016

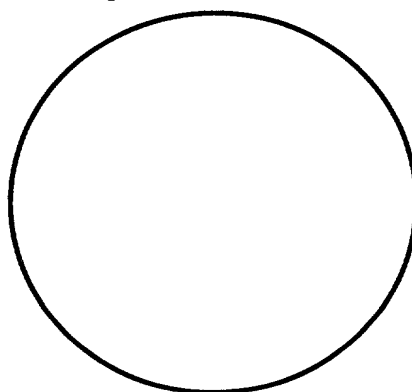
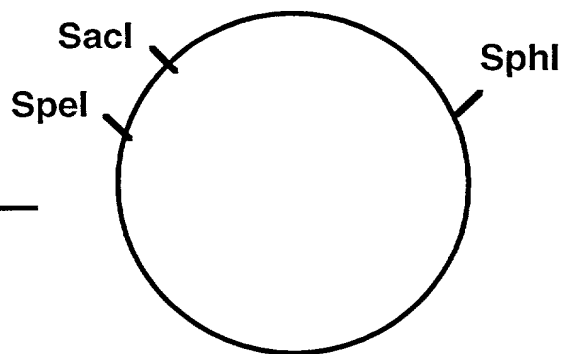
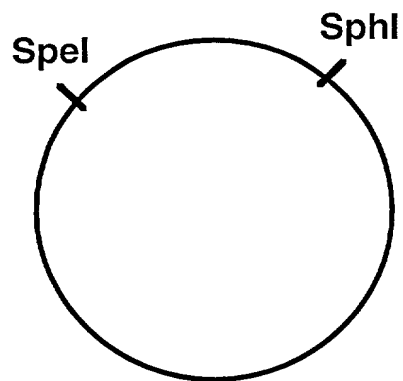


FIG. 8

pST182

pMP982



digest with **Spel**
and **SphI**, isolate
large frag.

digest with **Spel**
and **SacI**, isolate
647 bp frag.

digest with
SacI and **SphI**,
isolate 1809 bp
frag.



ligate

pMP1086

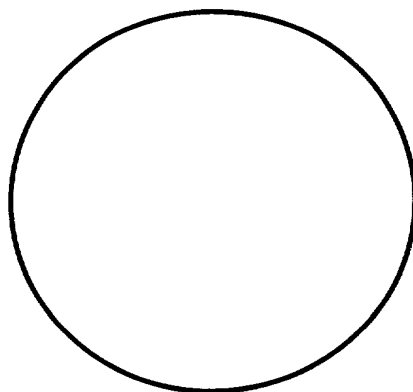
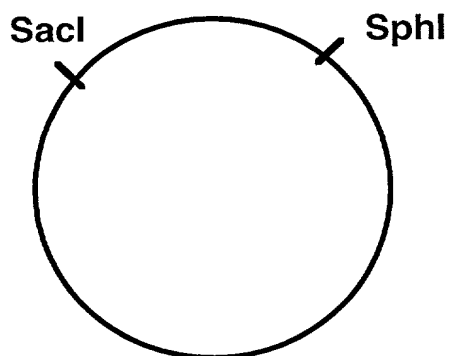


FIG. 9

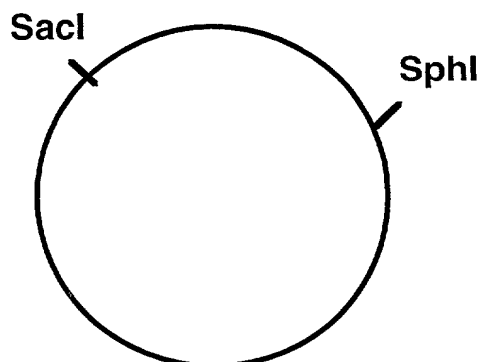
pMP1016



digest with
SacI and SphI,
isolate vector
frag.



pMP591



digest with
SacI and SphI,
isolate 1020 bp
frag.



ligate

pMP1099

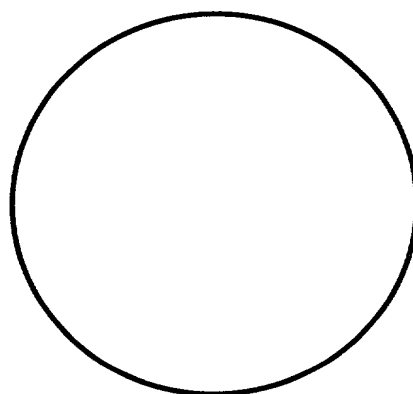
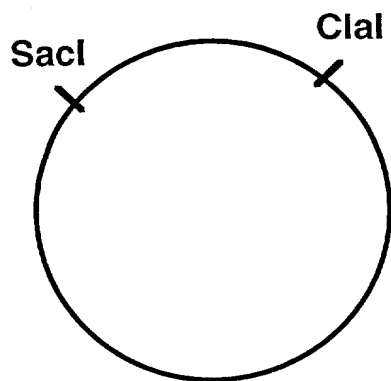
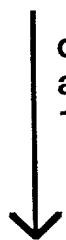


FIG. 10

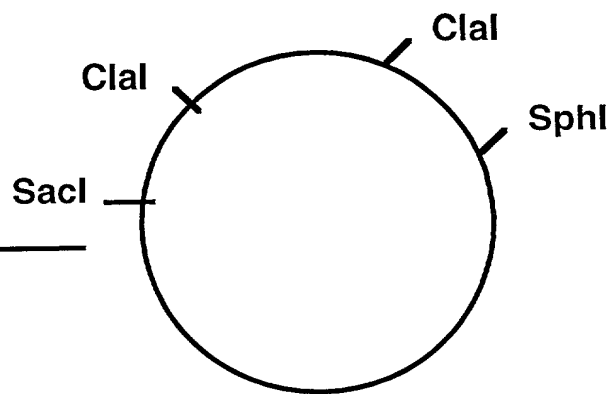
pMP945



digest with **SacI**
and **ClaI**, isolate
1007 bp frag.



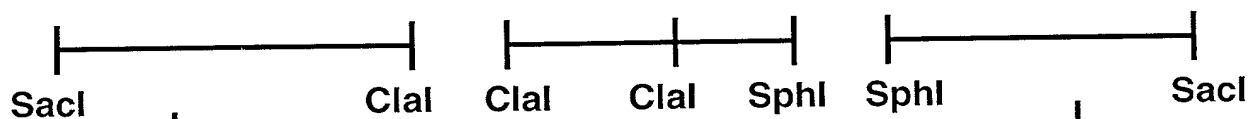
pMP1086



complete digest with
SphI, partial digest
with **ClaI**, isolate
814 bp frag.



digest with
SacI and **SphI**,
isolate large
frag.



ligate

pMP1201

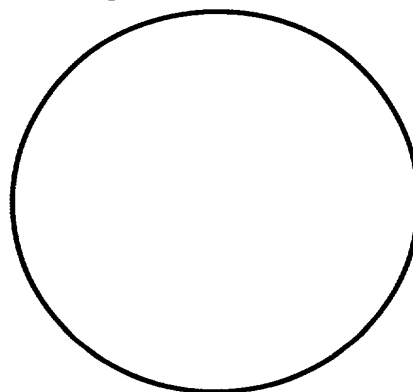
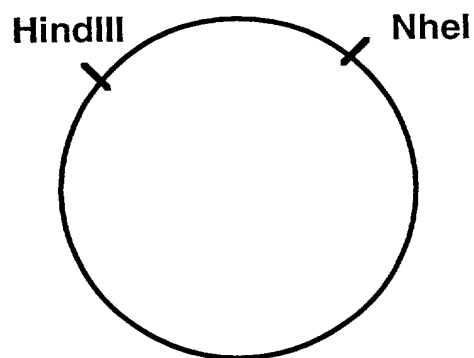


FIG. 11

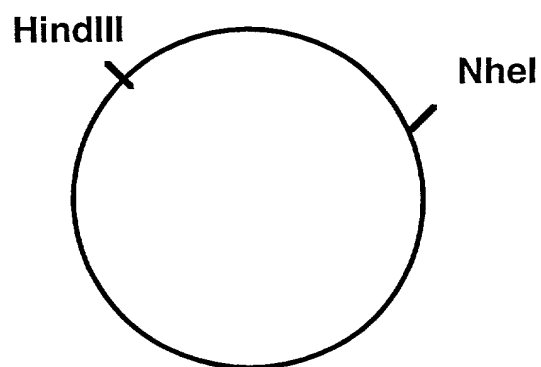
pMP951



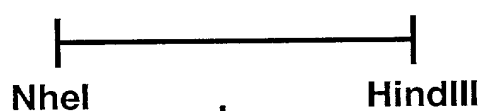
digest with HindIII
and NheI, isolate
large frag.



pMP945



digest with HindIII
and NheI, isolate
696 bp frag.



ligate

pMP1217

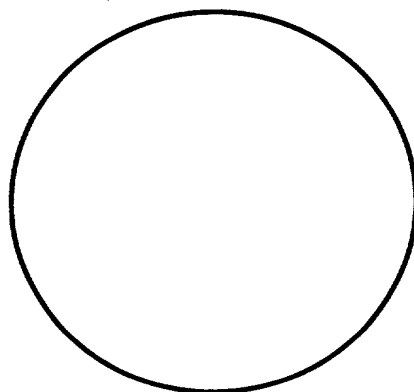
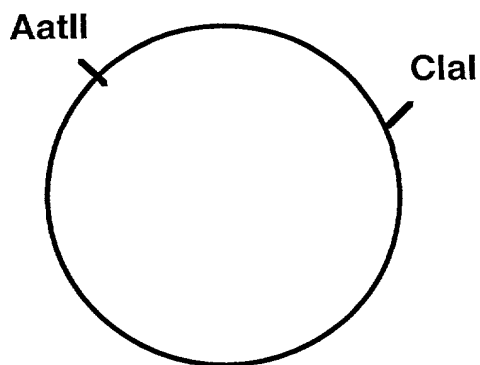
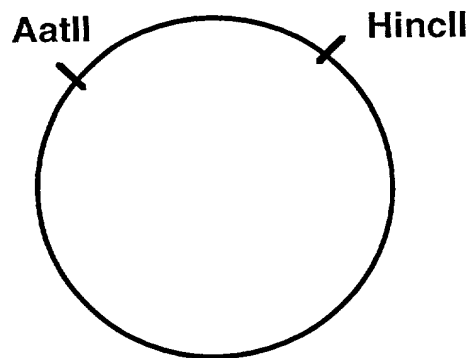


FIG. 12

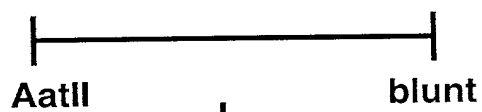
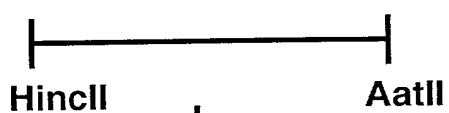
pACYC177

pJJ41



digest with AatII
and HincII, isolate
large frag.

digest with ClaI,
fill in with DNA
polymerase,
digest with AatII,
isolate 1021 bp
frag.



ligate

pJJ142

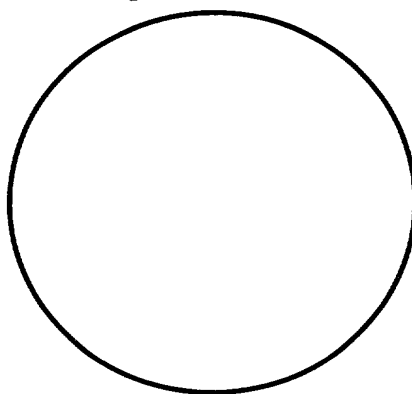
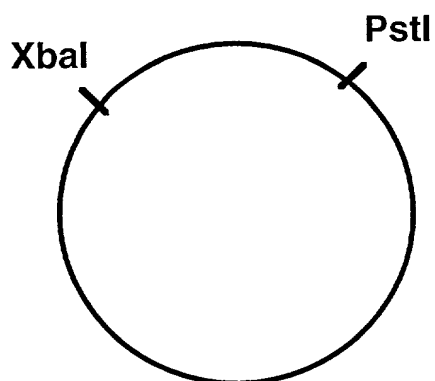
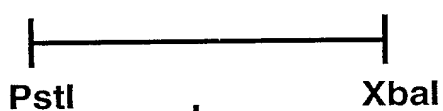


FIG. 13

pJJ142

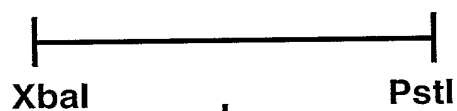


digest with XbaI and PstI, isolate large frag.



PCR reaction

digest PCR reaction with XbaI and PstI, isolate 488 bp frag.



ligate

pDR1

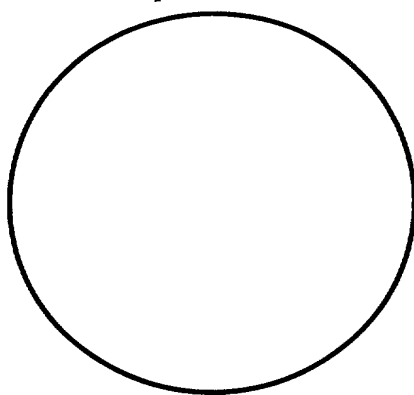
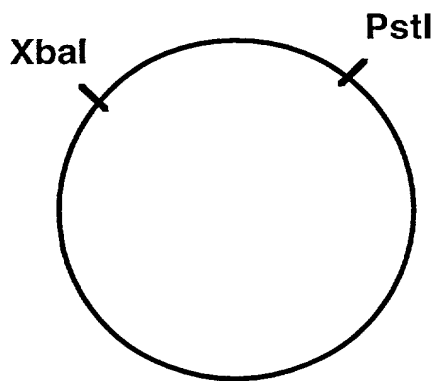


FIG. 14

pJJ142

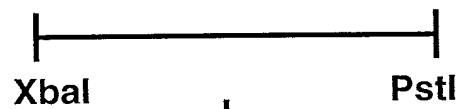


digest with XbaI and PstI, isolate large frag.



PCR reaction

digest PCR reaction with XbaI and PstI, isolate 491 bp frag.



ligate

pDR3

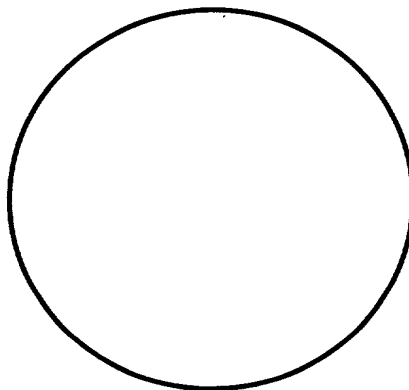


FIG. 15

1000 800 600 400 200 100 50 25 10 5 3 1

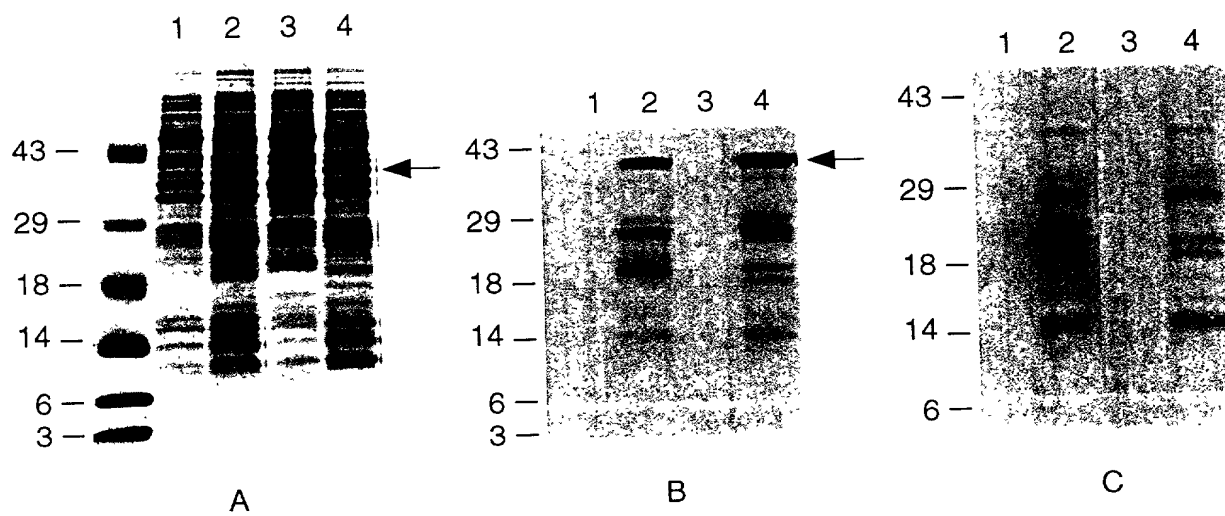


FIG. 16

trp	nutL site		Shine-
promoter	<hr/>		Dalgarno
-10	+1	A box	Met
		B box	
	<hr/>		
A TTAAGTAGTACGCAACGCTCTTACACATTCCAGCCCTGAAAAAAGGGCAAGTTACGTAAAAAGGATATCTAGAATTATG			

(SEQ ID NO:1)

AP

promoter +1

-10

A box B box

TATAGTCGCTTTGTTTATTTTTTAAATGTAATTGTAACTAGTAGCGAACGCTCTTACACATTCAGCCCTGAAAAAGGGCA-

(SEQ ID NO. 2)

Shine-
Dalgarno
Met

FIG. 17

1
 Tac II promoter +1
 -10 TTTAATGTGTGGAATTGTGAGCGGATAACAATTAAAGCTTTTATGGATGCACAAACA
 Shine-Dalgarno
 Start of λ N protein
 MetAspAlaGlnThr (SEQ ID NO: 3)
 (SEQ ID NO: 4)

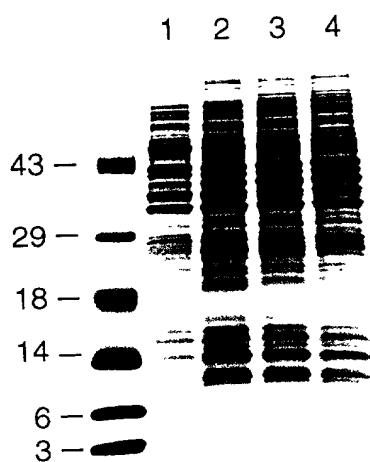
A

Tac II promoter +1
 -10 TTTAATGTGTGGAATTGTGAGCGGATAACAATTAAAGCTTAGGATTCTAGAAATTATGGATGCACAAACA
 Shine-Dalgarno
 Start of λ N protein
 MetAspAlaGlnThr (SEQ ID NO: 3)
 (SEQ ID NO: 5)

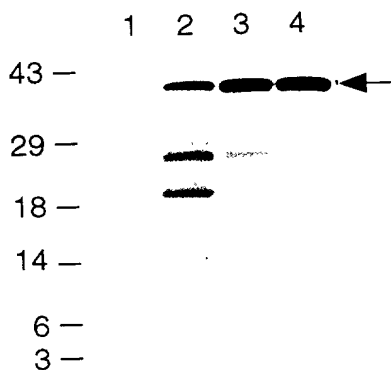
B

FIG. 18

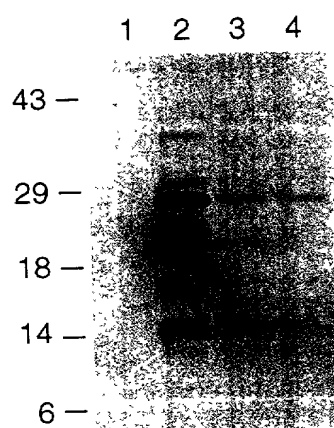
43 —
29 —
18 —
14 —
6 —
3 —



A

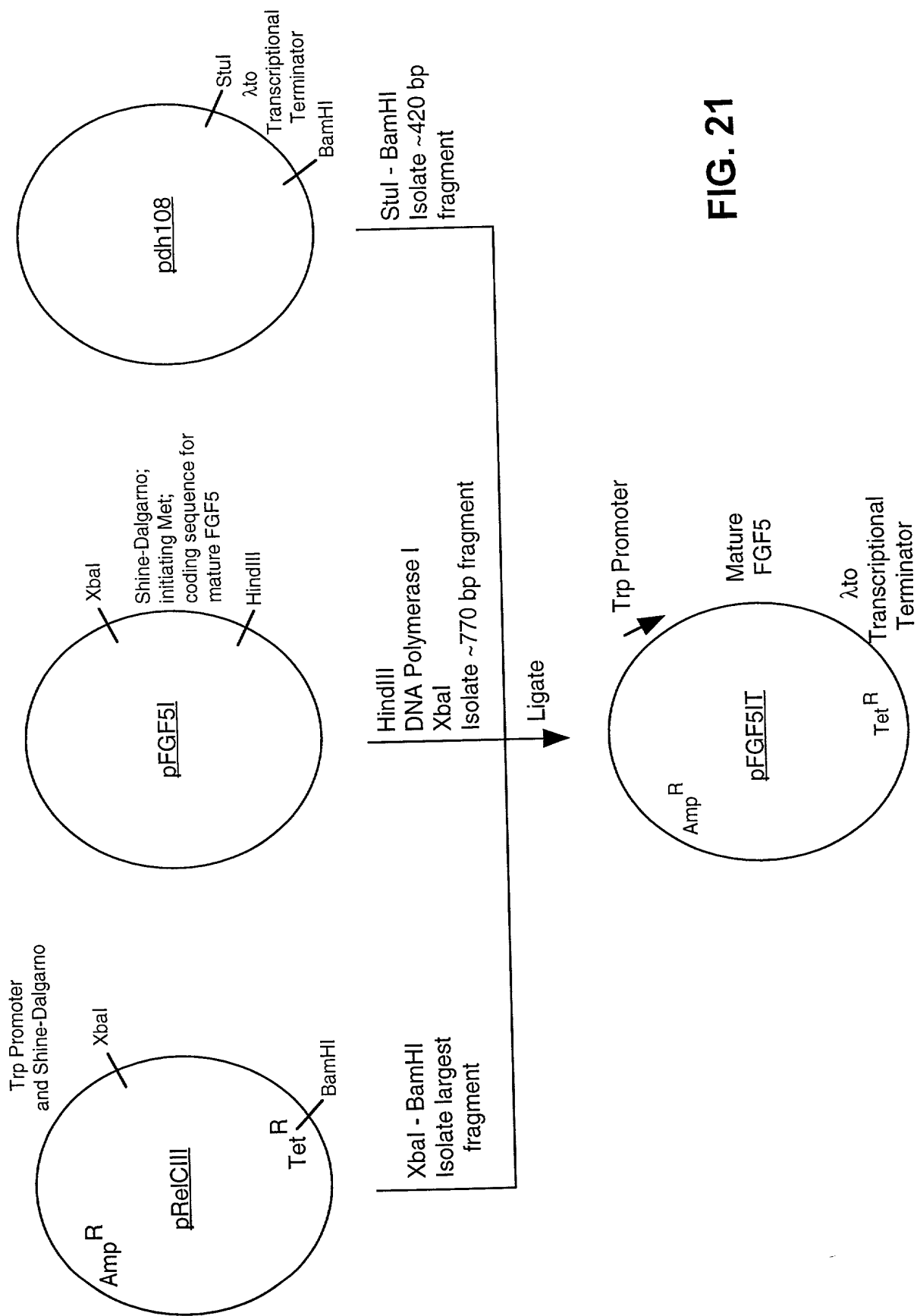


B



C

FIG. 19



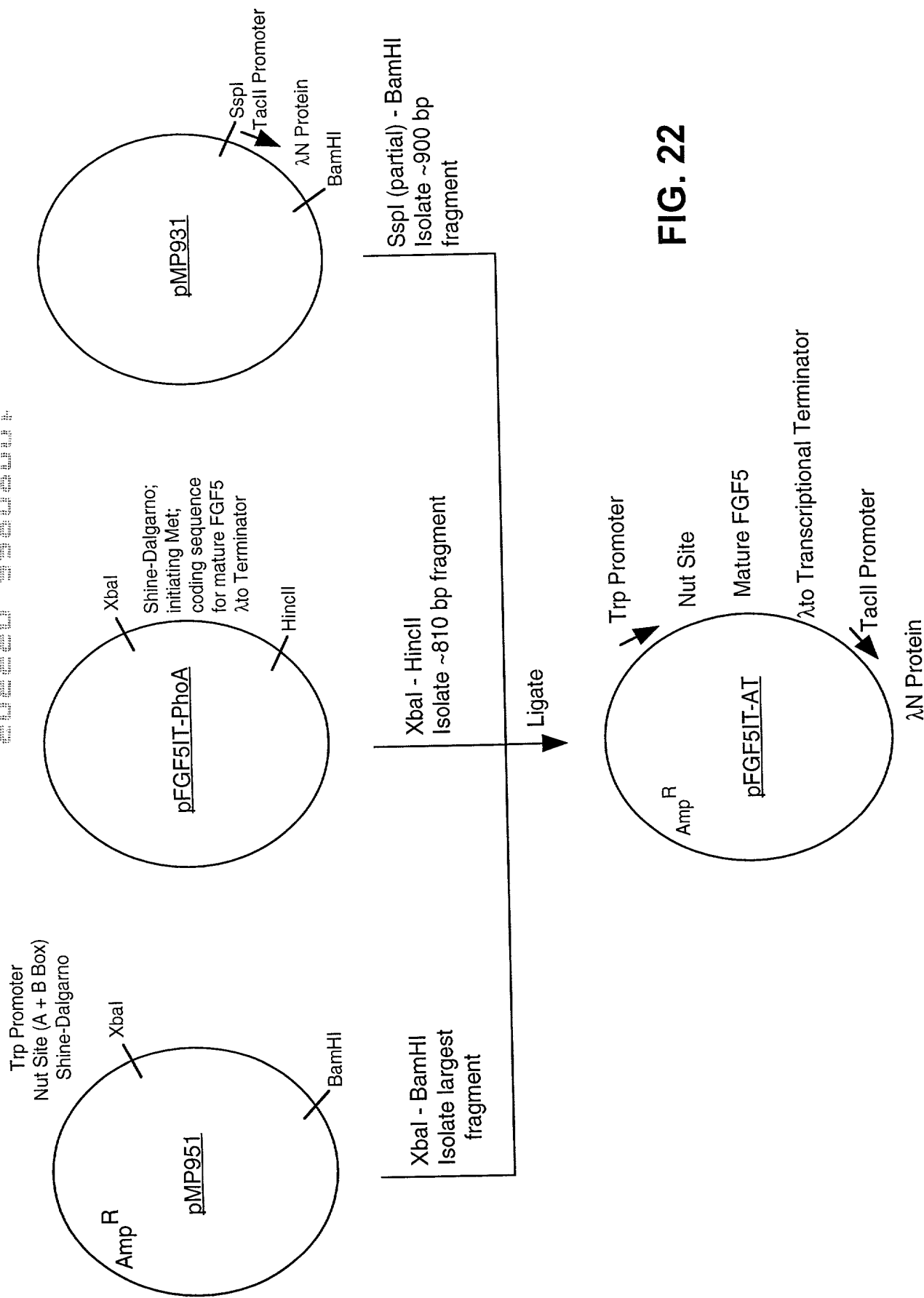


FIG. 22

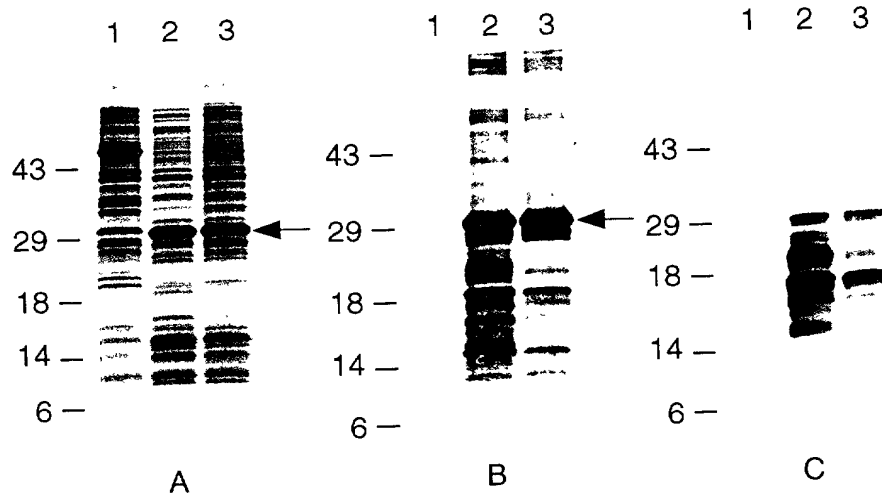


FIG. 23